FIG.1

[SEQ. ID NO: 3] X-C-C-T-T-G-A-G-A-T-T-T-C-C-C-T-C 5'

G-G-A-A-C-T-C-T-A-A-A-G-G-G-A-G-X
3' [SEQ. ID NO: 4]



X-C-C-T-T-G-A-G-A-T-T-T-C-C-C-T-C G-G-A-A-C-T-C-T-A-A-G-G-G-A-G-X

FIG.2

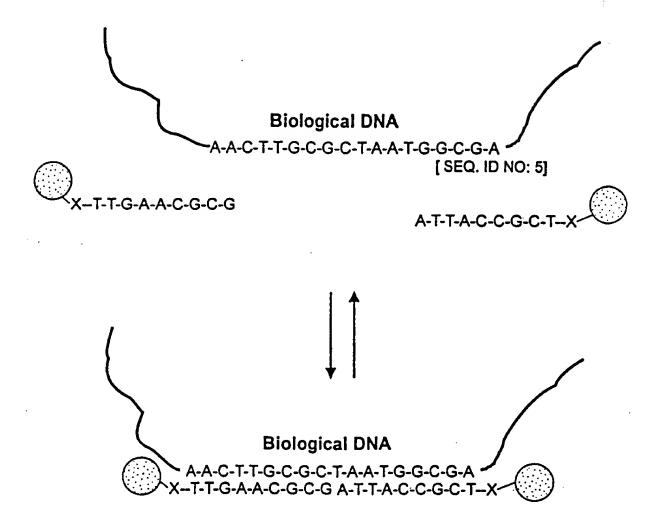
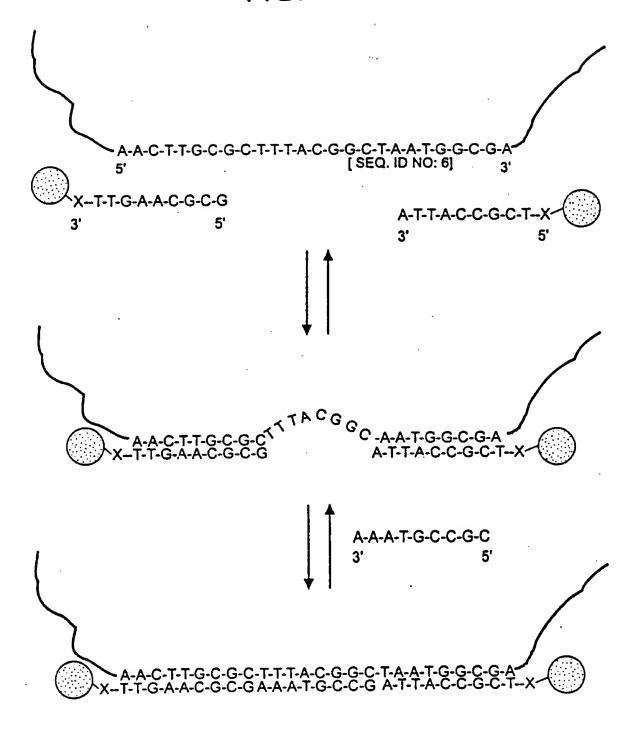
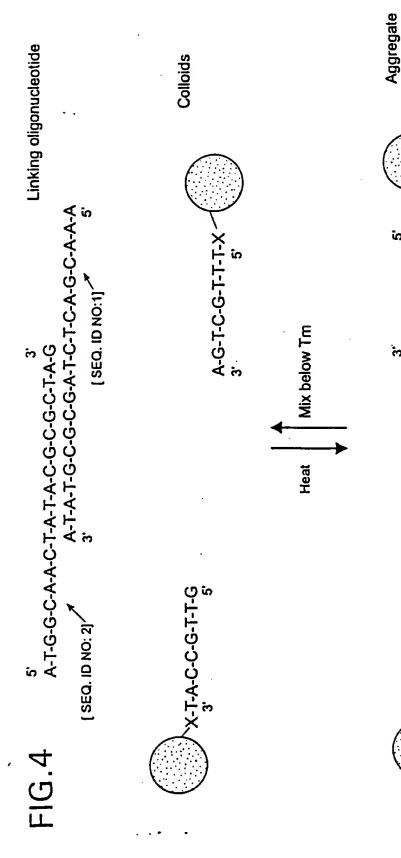


FIG.3





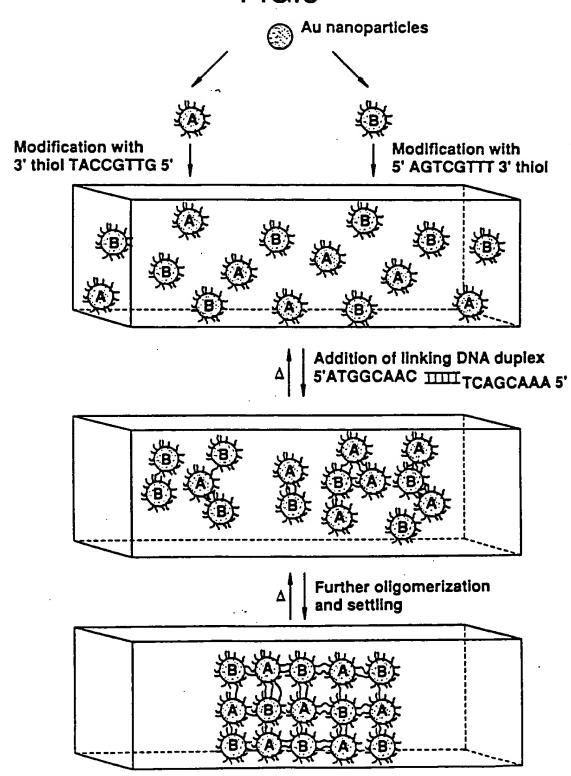
Precipitate (formed by further cross-linking)

Stand below Tm

Heat

A-T-G-G-C-A-A-C-T-A-T-A-C-G-C-G-C-T-A-G A-G-T-C-G-T-T-T-X X-T-A-C-C-G-T-T-G A-T-A-T-G-C-G-C-G-A-T-C-T-C-A-G-C-A-A-A 3'

FIG.5



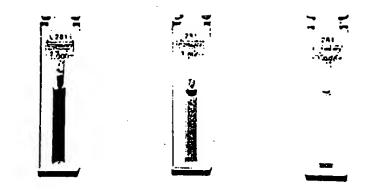
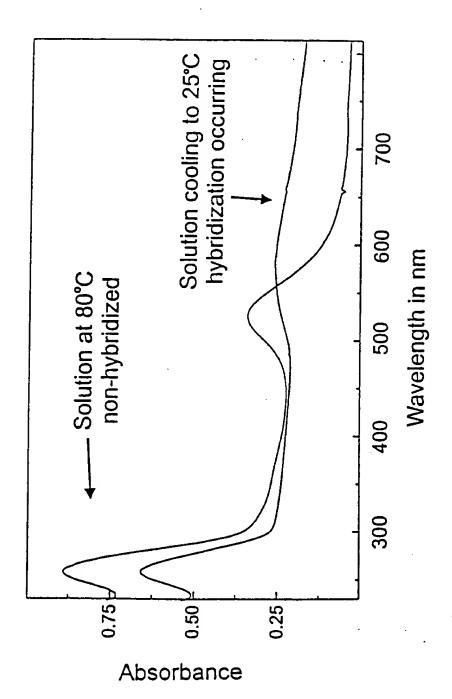
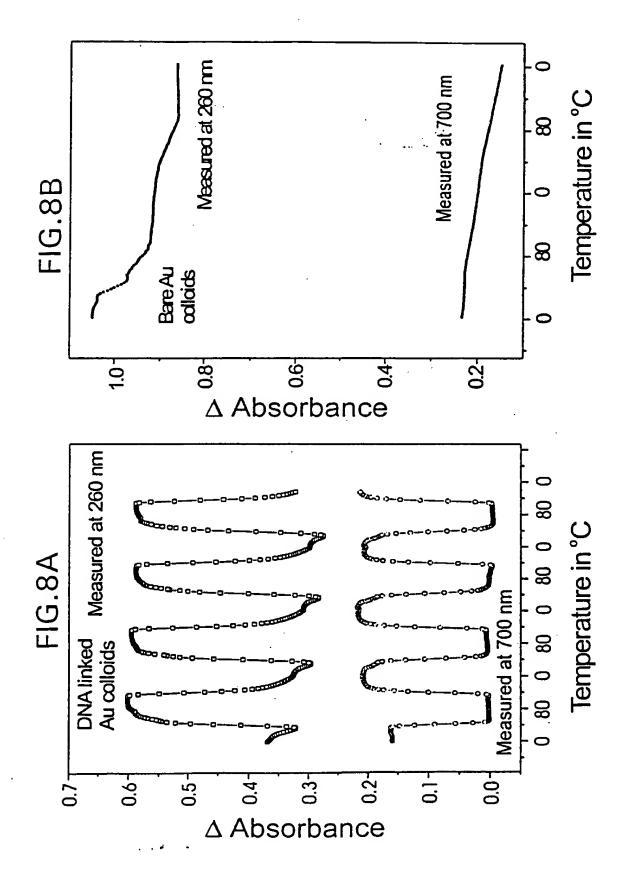


FIG.6A FIG.6B FIG.6C

FIG. 7





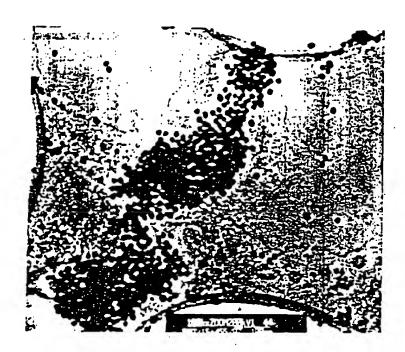


FIG.9A

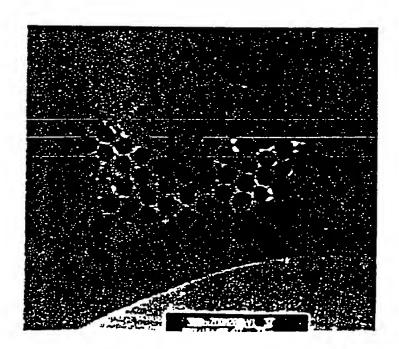
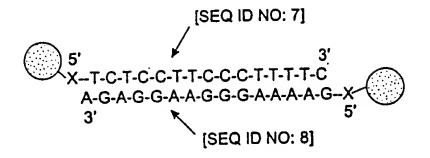
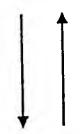


FIG.9B

FIG.10



3' T-C-T-C-C-T-T-C-C-C-T-T-T-C 5' [SEQ ID NO: 9]



5' 3' X--T-C-T-C-C-T-T-C-C-C-T-T-T-C A-G-A-G-G-A-A-G-G-A-A-A-G-X T-C-T-C-C-T-T-C-C-C-T-T-T-C 5'

FIG.11

_[SEQ. ID NO: 10]

S-A-T-G-G-C-A-A-C-T-A-C-G-C-G-C-T-A-G-A-G-T-C-G-T-T-T

T-A-C-C-G-T-T-G-A-T-A-T-G-C-G-C-G-A-T-C-T-C-A-G-C-A-A-S-7 5' 3' [SEQ. ID NO: 11]



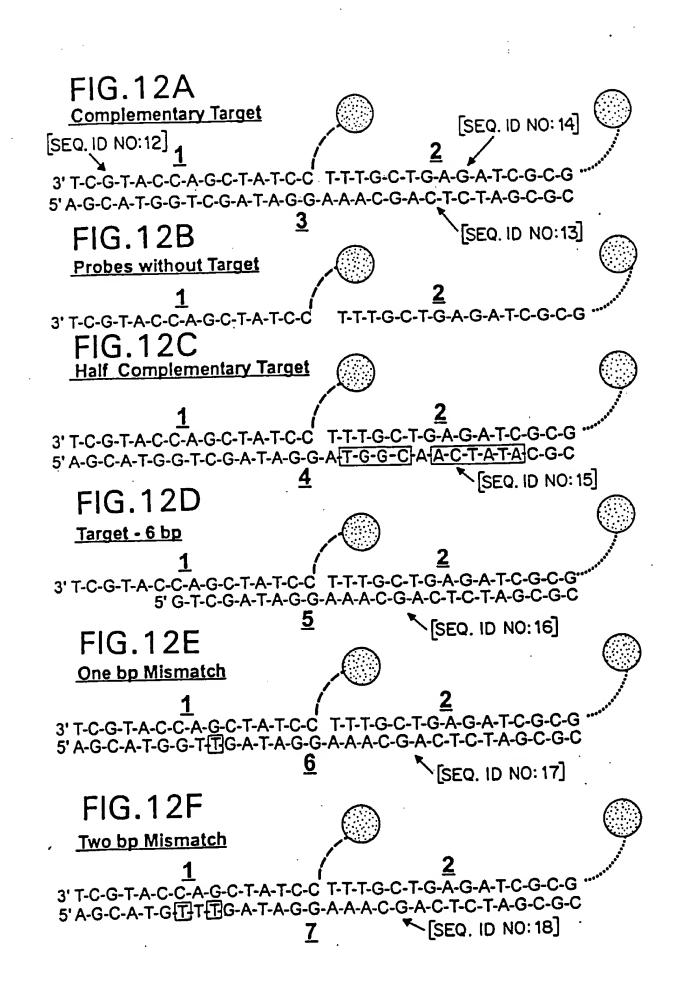


FIG.13A

transparent substrate

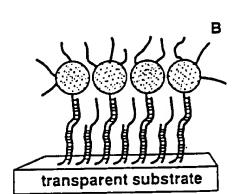
Modified DNA chemisorbed onto solid substrate

Analyte DNA hybridized onto substrate

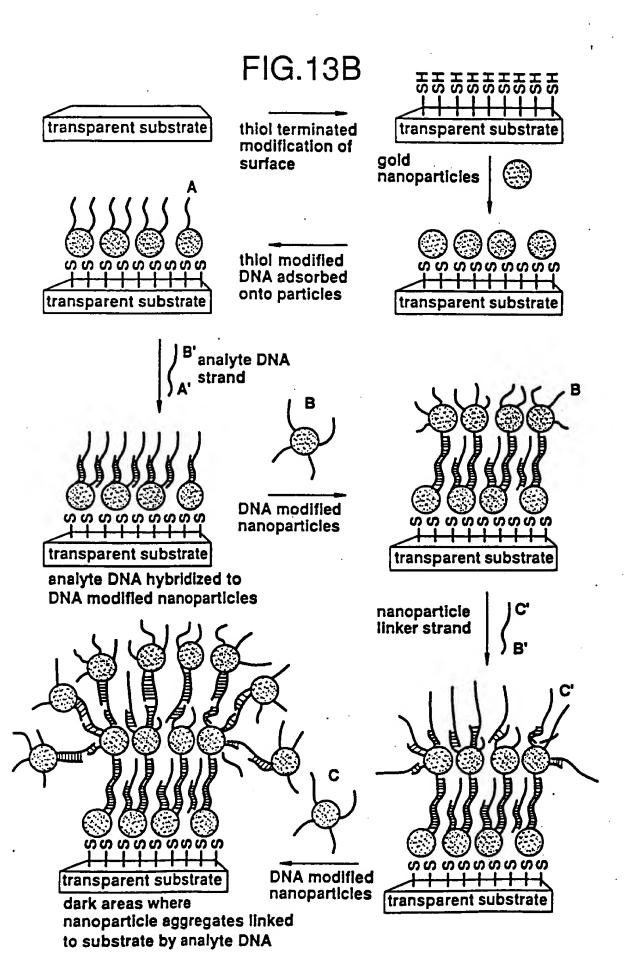
DNA modified colloids

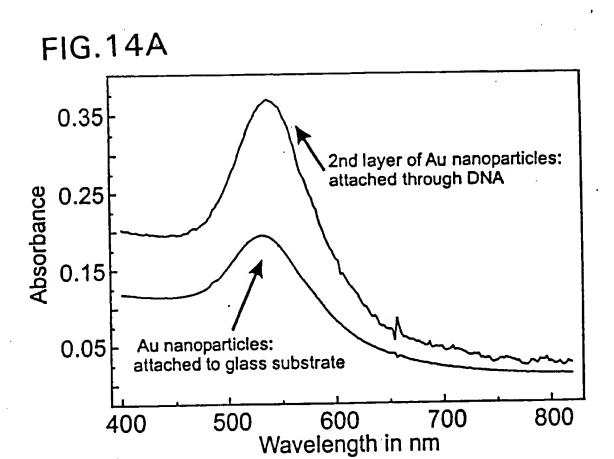
transparent substrate

Dark areas where nanoparticle aggregates are linked to substrate surface by analyte DNA



DNA modified colloids hybridized to bound analyte DNA





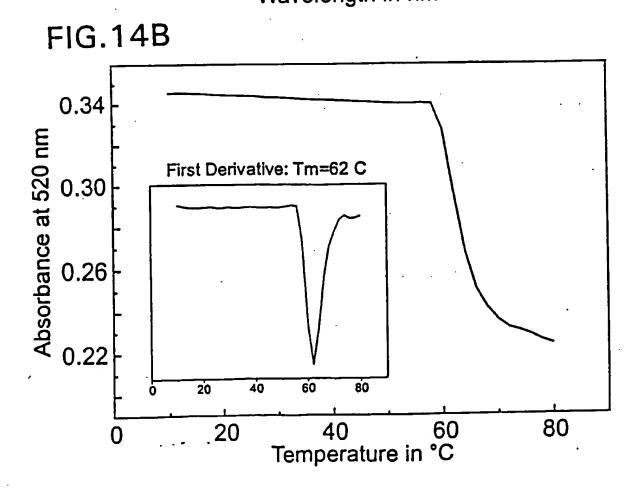


FIG15A Probes with No Target SEQ ID NO:19	
SEQ ID NO	:20
S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S)
FIG15B	
Half-Complementary Target	
3 SEQ ID NO:21	
5' TAC-GAG-TTG-AGA- <u>GAG-TGC-CCA-CAT</u> 3'	
S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S)
$\frac{1}{2}$	9
FIG15C	
Complementary Target Tm=53.5°C	
SEQ II	D NO:22
5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GCG 3'	
S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S	
<u>1</u> 2	
FIG15D	
I	4°C
0,12,000,10,1,1,1,1,1,1,1,1,1,1,1,1,1,1,	
5 SEQ ID No	0:23
5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC <u>T</u> 3'	
S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S	
S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S	
<u> </u>	
FIG15E Tm=46.2°C	
FIG15E Tm=46.2°C ONE Base-Pair Mismatch at Probe Tail	
FIG15E ONE Base-Pair Mismatch at Probe Tall 6 SEQ1	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3'	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3'	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 2 EIG15E	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15F Tm=51.6°C	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15F ONE Base Deletion	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15F ONE Base Deletion 7 SEQ ID NO:25	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15F ONE Base Deletion 7 SEQ ID NO:25 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC 3'	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S ONE Base Deletion 7 SEQ ID NO:25 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15F ONE Base Deletion 7 SEQ ID NO:25 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 2 1 2	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S ONE Base Deletion Tm=51.6°C ONE Base Deletion 7 SEQ ID NO:25 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 2 FIG15G	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S ONE Base Deletion 7 SEQ ID NO:25 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15F ONE Base Deletion 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15G ONE Base-Pair Insertion Tm=50.2°C	D NO:24
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S ONE Base Deletion Tm=51.6°C ONE Base Deletion 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GCD 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15G ONE Base-Pair Insertion Tm=50.2°C	
FIG15E ONE Base-Pair Mismatch at Probe Tail 5' TAC-GAG-TTG-AGA-CTC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S ONE Base Deletion 7 SEQ ID NO:25 5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GC 3' S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S 1 FIG15G ONE Base-Pair Insertion 8 Tm=50.2°C	

FIG. 16A 24 Base Template

5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GCG 3'

-- S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S --

48 Base Template with Complementary 24 Base Filler FIG. 16B

5' TAC-GAG-TTG-AGA-CCG-TTA-AGA-CGA-GGC-AAT-CAT-GCA-ATC-CTG-AAT-GCG 3' S-ATG-CTC-AAC-TCT GGC-AAT-TCT-GCT-CCG-TTA-GTA-CGT TAG-GAC-TTA-CGC-S

72 Base Template with Complementary 48 Base Filler FIG. 16C

5' TAC-GAG-TTG-AGA-CCG-TTA-AGA-CGA-GGC-AAT-CAT-GCA-TAT-ATT-GGA-CGC-TTT-ACG-GAC-AAC-ATC-CTG-AAT-GCG 3'

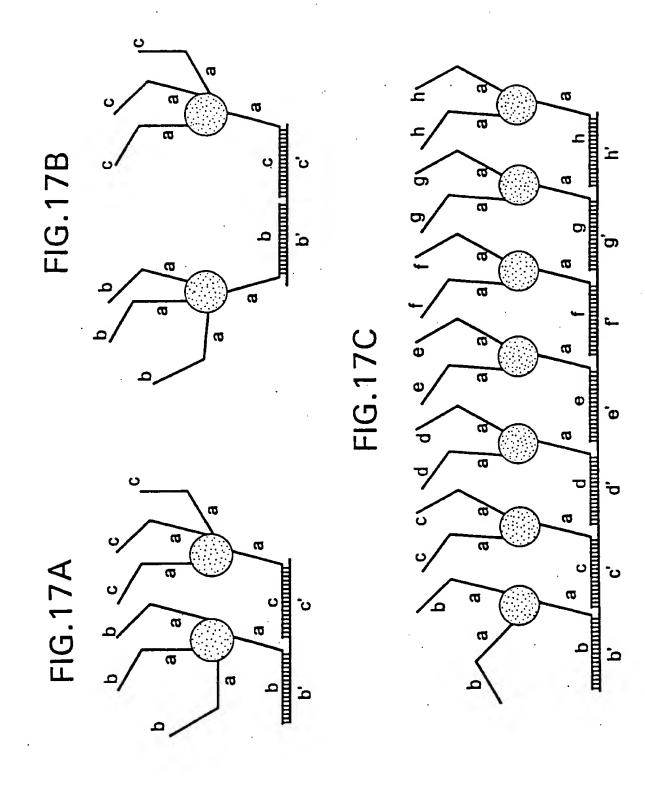
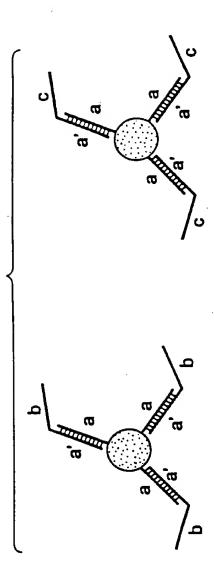
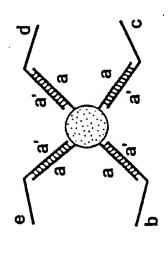


FIG.17D





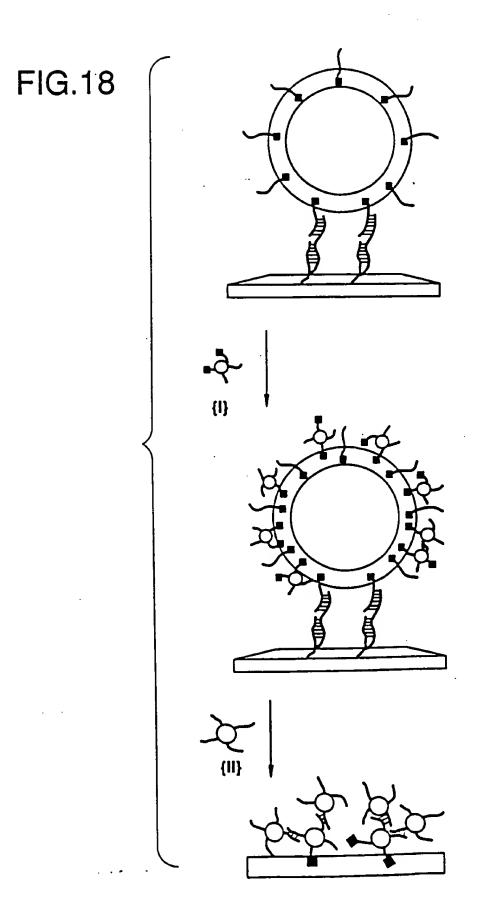
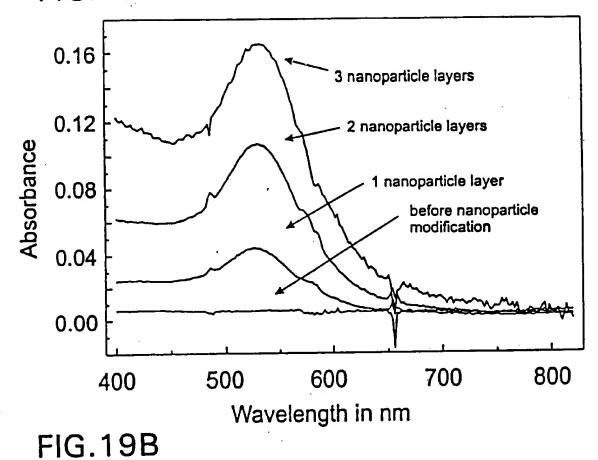


FIG.19A

0.16



0.24 Absorbance at 520 nm First Derivative: Tm=55°C 0.20

80

40

Temperature in °C

80

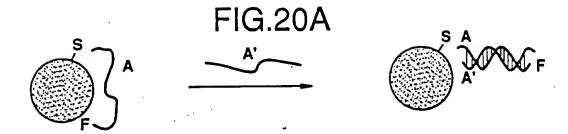
60

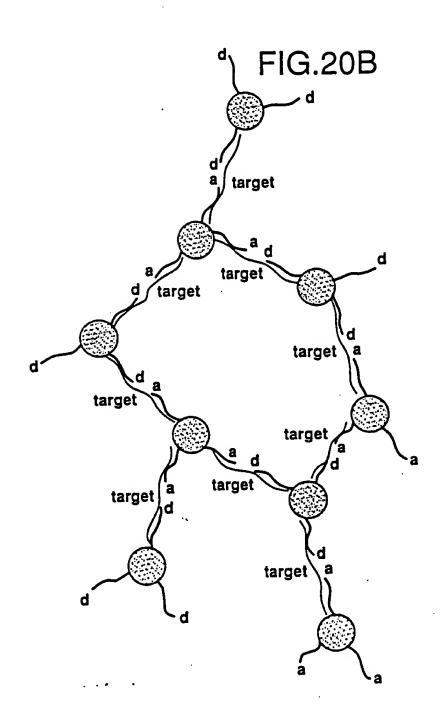
40

20

20

60





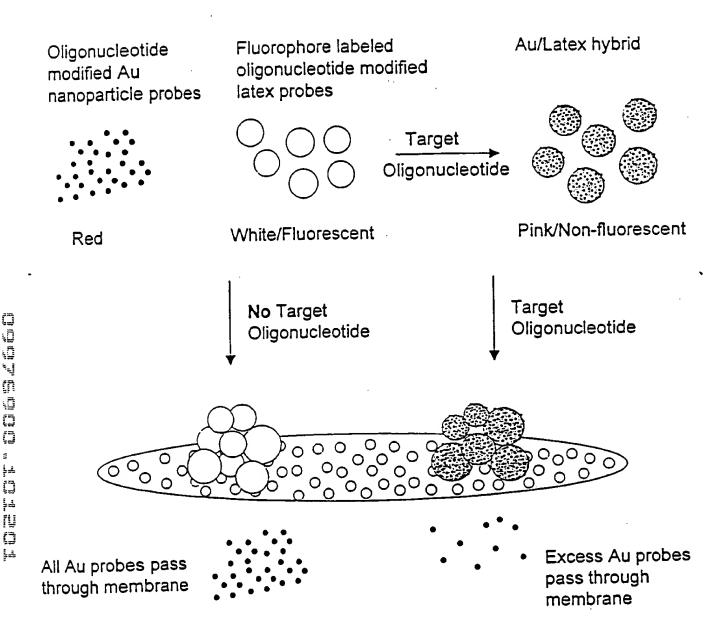
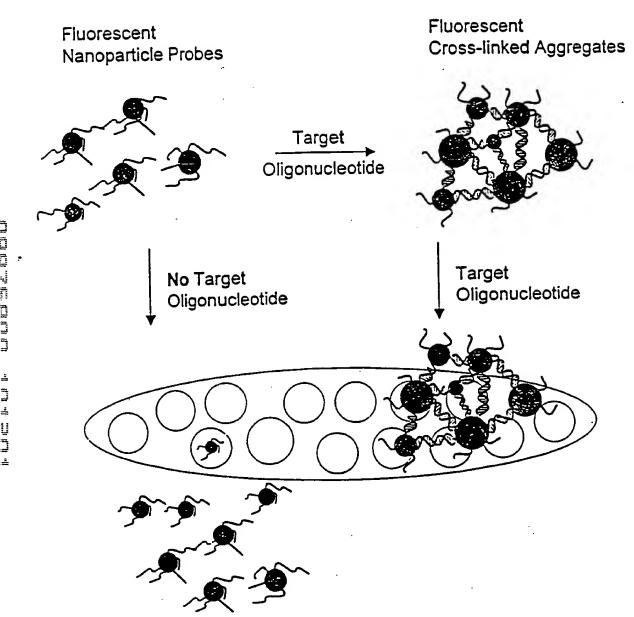


FIGURE 21



The fluorescent nanoparticle probes pass through the membrane

The fluorescent cross-linked aggregates are retained by the membrane

Anthrax PCR Product

5'G GCG GAT GAG TCA GTA GTT AAG GAG GCT CAT AGA GAA GTA ATT AAT 3'C CGC CTA CTC AGT CAT CAA TTC CTC CGA GTA TCT CTT CAT TAA TTA

TCG TCA ACA GAG GGA TTA TTG TTA AAT ATT GAT AAG GAT ATA AGA AAA AGC AGT TGT CTC CCT AAT AAC AAT TTA TAA CTA TTC CTA TAT TCT TTT

ATA TTA TCC AGG GTT ATA TTG TAG AAA TTG AAG ATA CTG AAG GGC TT 3'
TAT AAT AGG TCC CAA TAT AAC ATC TTT AAC TTC TAT GAC TTC CCG AA 5'

141 mer Anthrex PCR product [SEQ 10 NO:36]

3' CTC CCT AAT AAC AAT-

[SEQ 10 NO:31]

3' TTA TAA CTA TTC CTA LSED ID NO: 38]

Oligonucleotide-Nanoparticle Probes

Blocker Oligonucleotides

3' C CGC CTA CTC AGT CAT CAA TTC CTC CGA GT [SEQ 15 ND: 39]

3' A TCT CTT CAT TAA TTA AGC AGT TGT [SEQ 15 ND: 40]

3' TAT TCT TTT TAT AAT AGG TCC CAA TAT [SEQ 15 ND: 41]

3' AAC ATC TTT AAC TTC TAT GAC TTC CCG AA [SEQ 16 ND: 42]

FIGURE 23

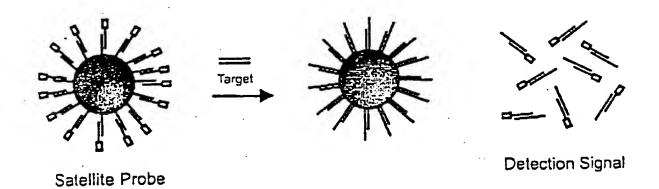
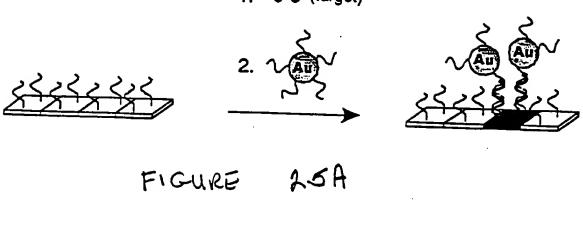
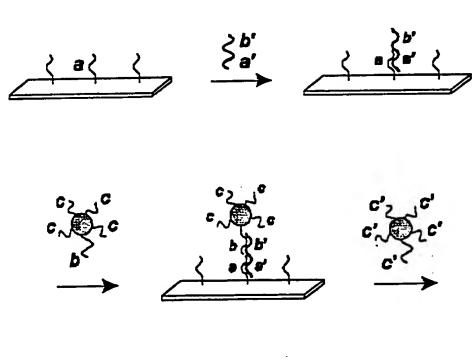


FIGURE 24

1. **(**target)





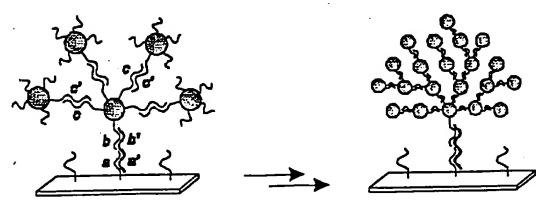


FIGURE 25 B

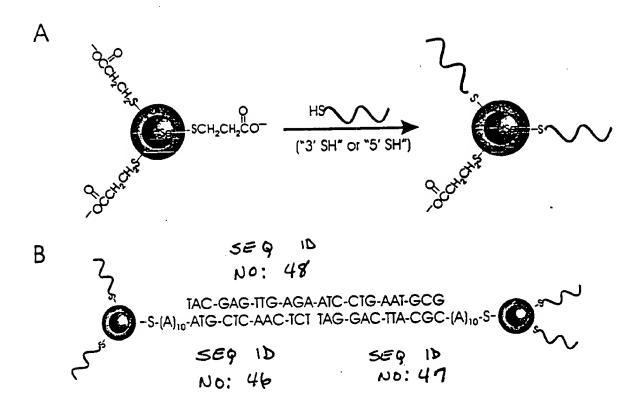
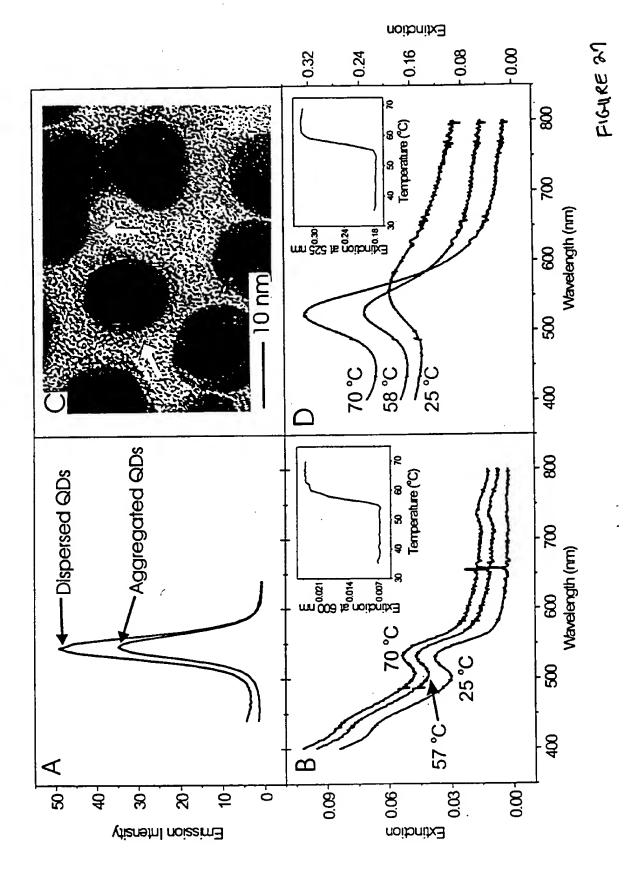


FIGURE 26

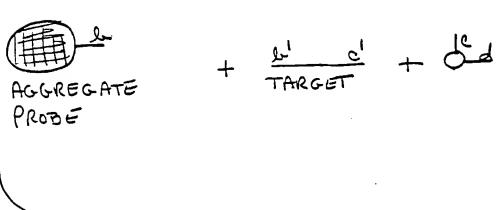


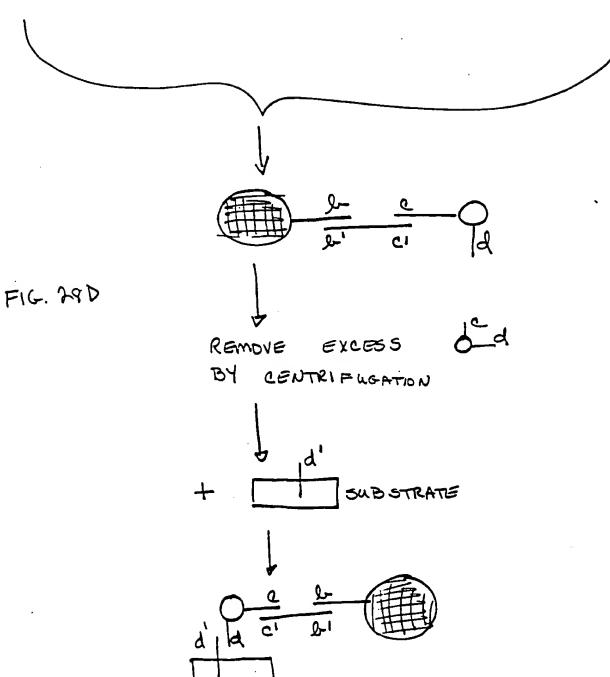
$$\frac{a + \frac{a'}{a'} + \frac{a'}{a'}}{a'} \rightarrow 0$$

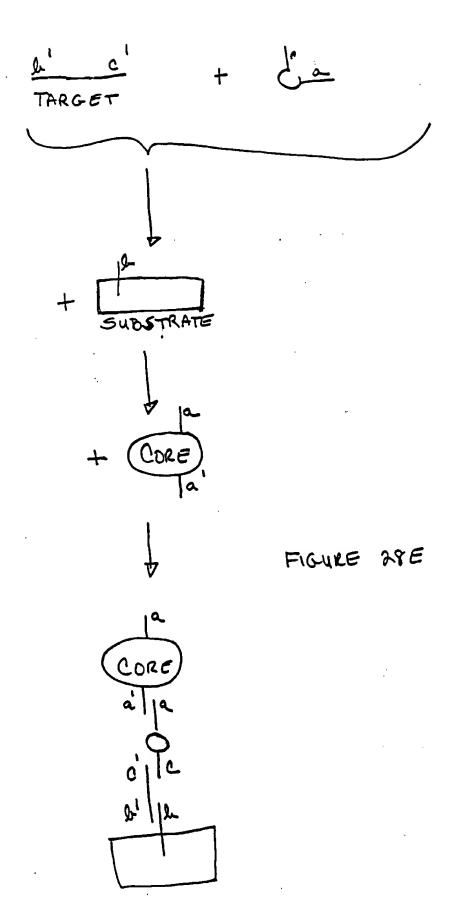
FIGURE Z8A

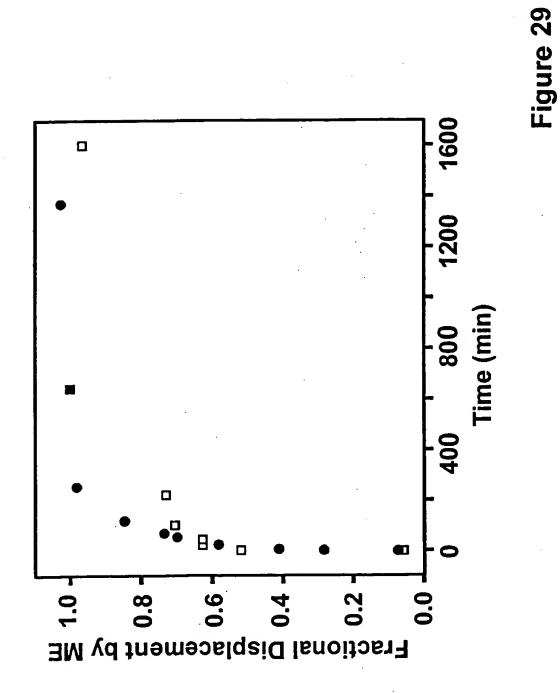
FIGURE AS B

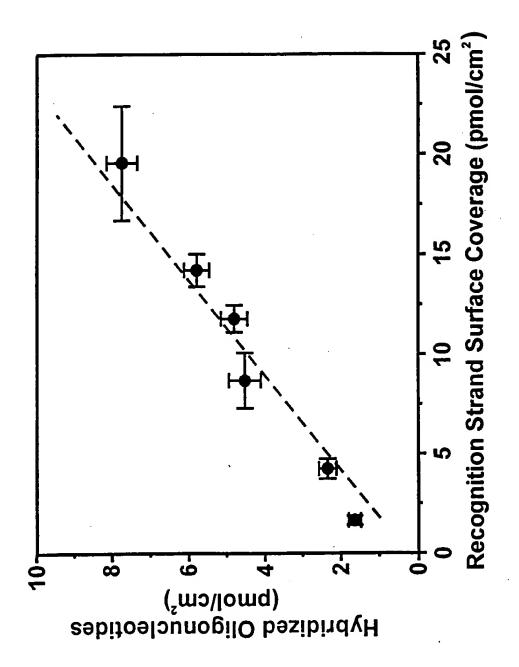
FIG. 28C

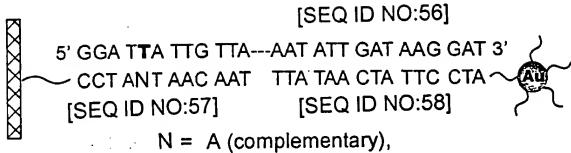












N = A (complementary), G,C,T (mismatched)

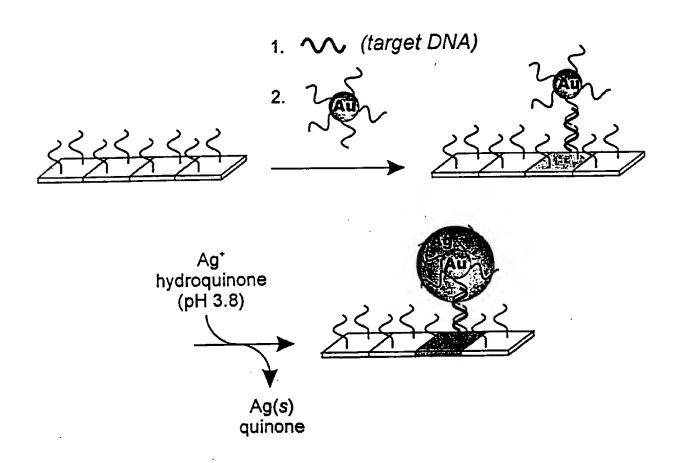


Figure 32

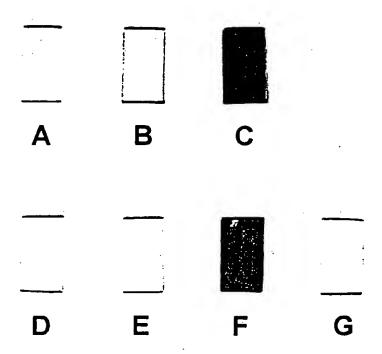


Figure 33

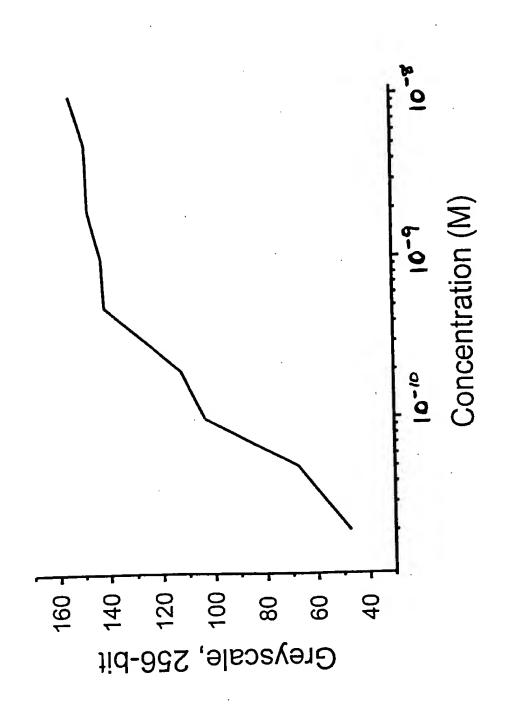
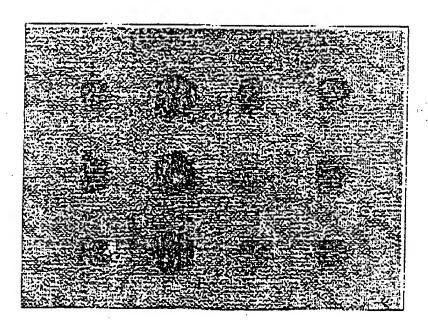


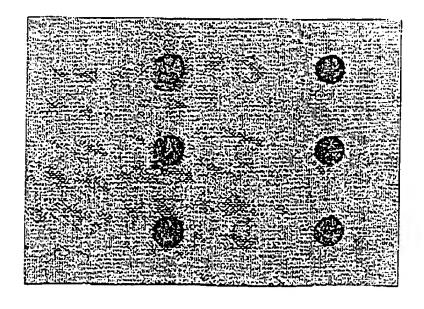
Figure 34

Figure 35

Fig. 364



F16.36B



C 🛕 T G

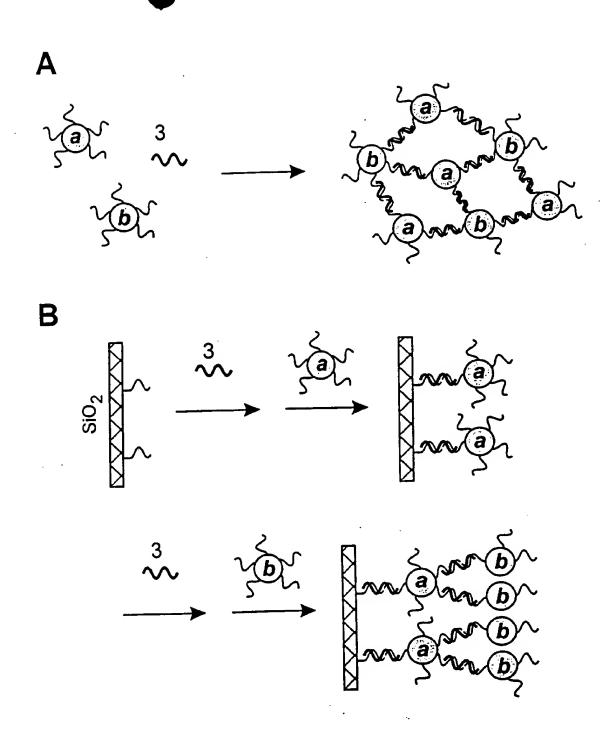


Figure 37

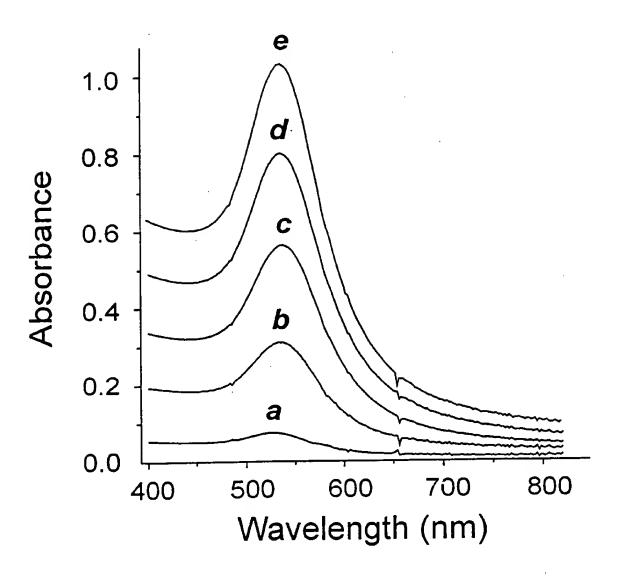


Figure 38A

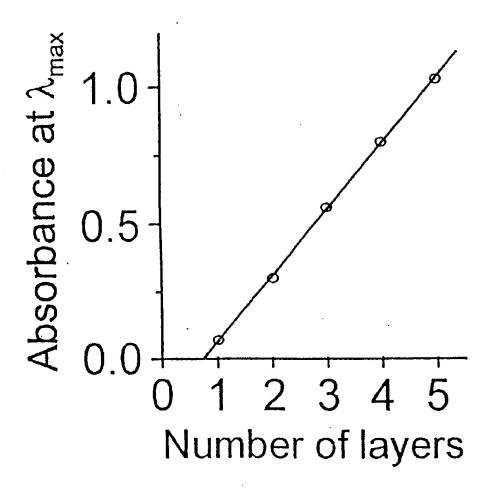


Figure 38B

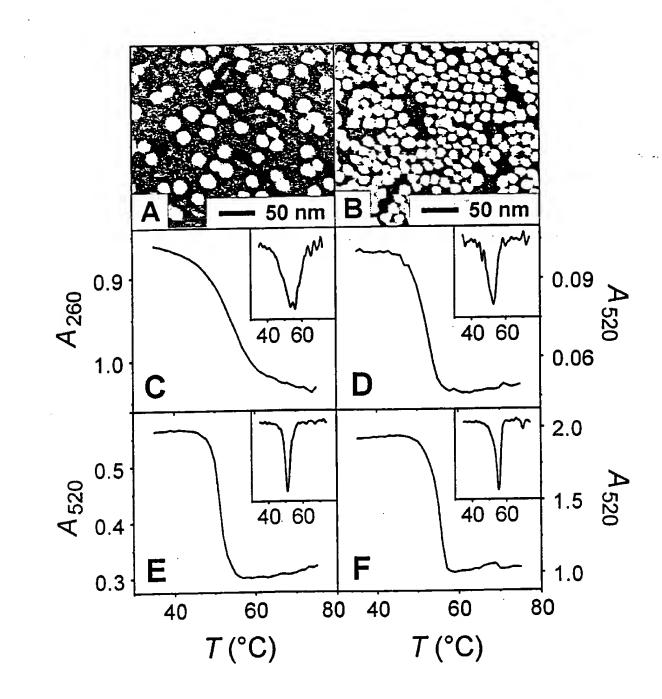
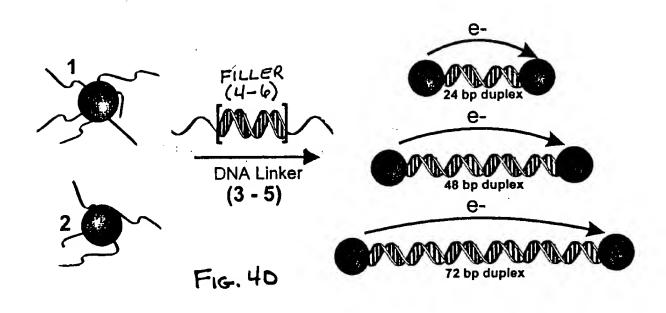


Figure 39



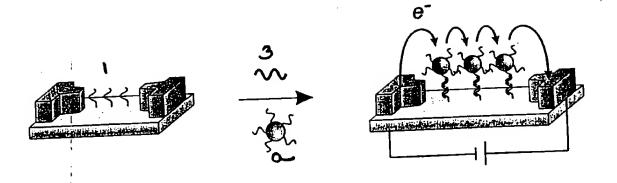


FIG. 41